AMENDMENTS

Claims:

Please amend the claims as indicated hereafter.

1. (Previously canceled)

2. (Previously Amended) The method according to claim 37, wherein the primer is a fragment of deoxyribonucleic or ribonucleic acid, an oligodeoxyribonucleotide, an oligoribonucleotide, or a copolymer of deoxyribonucleic acid and ribonucleic acid.

3. (Previously Amended) The method according to claim 37, wherein the nucleic acid of interest is deoxyribonucleic acid, a ribonucleic acid, or a copolymer of deoxyribonucleic acid and ribonucleic acid.

4. (Canceled).

5. (Previously Amended) The method according to claim 37, wherein the terminator nucleotide is a dideoxyribonucleotide or an analogue thereof and the non-terminator nucleotide is a deoxyribonucleotide or a ribonucleotide or an analogue thereof.

6. (Canceled).

7. (Canceled).

8. (Previously Amended) The method according to claim 37, wherein in step (d), the duplex from step (c) is contacted with non-terminator nucleotides, wherein each non-terminators is labeled with the same or different detectable marker.

9. (Previously Amended) The method according to claim 37, wherein said detectable

marker comprises an enzyme, radioactive isotope, a fluorescent molecule, or a protein ligand.

10. (Canceled)

11. (Previously Amended) The method according to claim 37, wherein said enzyme is template-dependent.

12. (Original) The method of claim 11, wherein the template-dependent enzyme is DNA polymerase.

13. (Original) The method according to claim 12, wherein the DNA of polymerase is *E. coli* DNA polymerase I or the "Klenow fragment" thereof, T4 DNA polymerase, T7 DNA polymerase, or *T. aquaticus* DNA polymerase.

14. (Original) The method according to claim 11, wherein said enzyme is RNA polymerase or reverse transcriptase.

15. (Previously amended) The method according to claim 37, wherein the primer comprises on or more moieties that permit affinity separation of the primer from the unincorporated reagent and/or the nucleic acid of interest.

16. (Previously amended) The method according to claim 37, wherein the primer comprises one or more moieties that links the primer to a solid surface.

17. (Original) The method according to claim 15, wherein the moieties comprises biotin or digitonin.

18. (Original) The method according to claim 16, wherein the moieties comprises biotin or digitonin.

19. (Original) The method according to claim 15, wherein the moieties comprises a DNA or RNA sequence that permits affinity separation of the primer from the unincorporated reagent and/or the nucleic acid of interest via base pairing to a complementary sequence present in a nucleic acid attached to a solid support.

20. (Original) The method according to claim 16, wherein the moieties comprises a DNA or RNA sequence that permits affinity separation of the primer from the unincorporated reagent and/or the nucleic acid of interest via base pairing to a complementary sequence present in a nucleic acid attached to a solid support.

21. (Original) The method according to claim 15, wherein the moieties comprises a DNA or RNA sequence that allows the primer to link to a solid support via base pairing to a complementary sequence present in solid surface.

22. (Original) The method according to claim 16, wherein the moieties comprises a DNA or RNA sequence that allows the primer to link to a solid support via base pairing to a complementary sequence present in solid surface.

23. (Previously amended) The method according to claim 37, wherein the nucleic acid of interest has been synthesized enzymatically *in vivo, in vitro,* or synthesized non-enzymatically.

24. (Previously amended) The method according to claim 37, wherein the nucleic acid of interest is synthesized by polymerase chain reaction.

25. (Previously Amended) The method according to claim 37, wherein the nucleic acid of interest comprises non-natural nucleotide analogs.

26. (Original) The method according to claim 25, wherein the non-natural nucleotide

analogs comprise deoxyinosine or 7-dezaz-2'-deoxyguanosine.

27. (Previously amended) The method according to claim 37, wherein the sample comprises genomic DNA from an organism, RNA transcript thereof, or cDNA prepared from RNA transcripts thereof.

28. (Previously Amended) The method according to claim 37, wherein the sample comprises extragenomic DNA from an organism, RNA transcripts thereof, or cDNA prepared from RNA transcripts thereof.

29. (Previously Amended) The method according to claim 27, wherein the organism is a plant, microorganism, bacteria, or virus.

30. (Previously Amended) The method according to claim 28, wherein the organism is a plant, microorganism, bacteria, or virus.

31. (Original) The method according to claim 27, wherein the organism is a vertebrate or invertebrate.

32. (Original) The method according to claim 28, wherein the organism is a vertebrate or invertebrate.

33. (Original) The method according to claim 27, wherein the organism is a mammal.

34. (Original) The method according to claim 28, wherein the organism is a mammal.

35. (Original) The method according to claim 27, wherein the organism is a human being.

36. (Presently amended) The method according to claim ~~27~~28, wherein the organism is a human being.

37. (Currently amended) A method for detecting or quantifying the presence of a nucleic acid of interest having a variant of a known ~~target~~ nucleotide base in a predetermined position of a known nucleic acid ~~in a sample by detecting a signal from a plurality of labeled nucleotides incorporated into a primer extension product~~ comprising:

(a) ~~selecting~~ obtaining a nucleic acid of interest having a target nucleotide base at [[a]] the predetermined position in a template of [[a]] the nucleic acid of interest~~, wherein the target nucleotide base is a mutant nucleotide base or a known wild-type nucleotide base~~;

(b) preparing an unlabeled primer complementary to a sequence immediately upstream of the target nucleotide base;

(c) treating ~~a~~ sample containing the nucleic acid of interest, if the nucleic acid is double-stranded, so as to obtain unpaired nucleotide bases spanning the predetermined position, or directly employing step (d) if the nucleic acid of interest is single-stranded;

(d) annealing the primer from (b) with the nucleic acid of interest from (c) to obtain a primer-nucleic acid duplex, wherein the target nucleotide base in the nucleic acid of interest is the first unpaired base immediately downstream of the 3' end of the primer;

(e) mixing the primer-nucleic acid duplex from (d) with a primer extension reaction reagent comprising: (i) three types of non-terminator nucleotides that are not complementarily matched to the known ~~wild-type target~~ nucleotide base in the predetermined position of the nucleic acid of interest, wherein at least one type of the non-terminator nucleotide

is labeled with a detectable marker; and optionally (ii) one type of terminator nucleotide that is complementarily matched to the known ~~wild-type target~~ nucleotide base in the predetermined position of the nucleic acid of interest, wherein the terminator nucleotide is not labeled;

(f) extending the primer extension reaction by enzymatic or chemical means to form a labeled primer extension product comprising a plurality of labeled non-terminator nucleotides, wherein a labeled primer extension product does not form when the target nucleotide base in the predetermined position of the nucleic acid of interest is the same as the known target nucleotide base in the predetermined position of the known nucleic acid ~~wild-type~~; and

(g) determining the presence of the ~~mutant~~ nucleic acid of interest having the variant target nucleotide base at the predetermined position in the nucleic acid of interest by detecting the presence of the labeled primer extension product, wherein detecting the labeled primer extension product is not based on size.

38. (Canceled)

39. (Currently amended) A method for detecting the presence of a nucleic acid having a variant of a known nucleotide base at a predetermined position, comprising:

providing a nucleic acid having a ~~known wild-type~~ target nucleotide base ~~or a mutant target nucleotide base~~ at [[a]] the predetermined position;

annealing a primer to the nucleic acid immediately 3' of the predetermined position;

extending the primer in ~~one~~ an extension reaction to form a labeled primer extension product using a reaction mixture comprising non-terminator nucleotides, wherein ~~at least one~~ the non-terminator ~~nucleotide is~~ nucleotides are not complementarily matched to the ~~mutant~~ known nucleotide base at the predetermined position ~~target nucleotide~~ and at least one non-terminator

nucleotide is labeled with a detectable marker; and wherein a labeled primer extension product does not form when the target nucleotide base is the same as the known nucleotide base at the predetermined position ~~wild-type~~; ~~and~~

detecting the presence of the labeled primer extension product; and

correlating the presence of the labeled primer extension product with the presence of ~~a mutant target~~ a nucleic acid having a variant of the known nucleotide base at the predetermined position in the nucleic acid.

40. (Currently amended) A method for detecting the presence of a ~~mutant nucleotide in a~~ nucleic acid having a known nucleotide at a predetermined position, comprising:

providing a nucleic acid sample having the nucleic acid with a ~~known wild-type~~ target nucleotide ~~base or a mutant target nucleotide base~~ at [[a]] the predetermined position;

annealing a primer to the nucleic acid immediately 3' of the predetermined position;

extending the primer to form a labeled primer extension product using a reaction mixture comprising non-terminator nucleotides~~, wherein at least one non-terminator nucleotide is~~ that are not complementarily matched to the known ~~wild-type target~~ nucleotide base at the predetermined position and at least one non-terminator nucleotide is labeled with a detectable marker; and wherein a labeled primer extension product does not form when the identity of the target nucleotide base ~~is mutant~~ is the same as the known nucleotide base at the predetermined position; and

detecting the presence of the labeled primer extension product, wherein the absence of a detectable ~~detection of the~~ labeled primer extension product ~~is not based on the size of the labeled extension product, and wherein detecting a labeled primer extension product~~ indicates the presence of ~~a wild-type~~ the known nucleotide base at the predetermined position in the nucleic

acid.

41. (Currently amended) A method for detecting or quantifying the presence of a known target nucleic acid in a sample by detecting a signal from a plurality of labeled nucleotides incorporated into a primer extension product comprising:

(a) selecting a nucleic acid having a target nucleotide base at a predetermined position in a template of a nucleic acid of interest~~, wherein the target nucleotide base is a known mutant nucleotide base or a known wild-type nucleotide base~~;

(b) preparing an unlabeled primer complementary to a sequence immediately upstream of the target nucleotide base;

(c) treating a sample containing the nucleic acid of interest, if the nucleic acid is double-stranded, so as to obtain unpaired nucleotide bases spanning the predetermined position, or directly employing step (d) if the nucleic acid of interest is single-stranded;

(d) annealing the primer from (b) with the nucleic acid of interest from (c) to obtain a primer-nucleic acid duplex, wherein the target nucleotide base in the nucleic acid of interest is the first unpaired base immediately downstream of the 3' end of the primer;

(e) mixing the primer-nucleic acid duplex from (d) with a primer extension reaction reagent comprising: (i) ~~at least one types of~~ non-terminator nucleotides that are not complementarily matched to the known ~~mutant~~ target nucleotide base, wherein at least one type of the non-terminator nucleotide is labeled with a detectable marker; and optionally (ii) one type of terminator nucleotide that is complementarily matched to the known ~~mutant~~ target nucleotide base, wherein the terminator nucleotide is not labeled;

(f) extending the primer extension reaction by enzymatic or chemical means to form a labeled primer extension product comprising a plurality of labeled non-terminator nucleotides,

wherein a labeled primer extension product does not form when the target nucleotide base is the known nucleotide ~~mutant~~; and

(g) wherein the absence of a detectable primer extension product indicates ~~determining~~ the presence of the known ~~wild-type~~ target nucleotide at the predetermined position in the nucleic acid of interest ~~by detecting the presence of the labeled nucleotide in primer extension product, wherein detecting the labeled nucleotide primer extension product is not based on size~~.

REMARKS

This is a full and timely response to the outstanding Final Office Action mailed September 23, 2003. Claims 2-9, 11-37 and 39-41 are pending. Reconsideration and allowance of the application and presently pending claims, as amended, are respectfully requested. Applicant submits that the amendments do not raise new issues or require additional searching because the amendments are clarifying amendments and do not introduce additional substantive elements.

1. Amendments to the Claims

By the present response, Applicant cancels claims 4, 6, and 7, and amends claims 37 and 39-41. Claims 37 and 39-41 are amended to replace the term "mutant" with --variant-- and to delete the term "wild-type." Basis for this amendment is found throughout the specification as originally filed, for example, p. 13, last full paragraph. Additionally conforming and clarifying amendments are provided to further clarify the scope of the claims. Applicant believes no new matter is introduced by these amendments.

2. Rejection of Claims 2-9, 11-37 and 39-41 Under 35 U.S.C. § 102(e)

The final Office Action maintains the rejection of claims 2-9 and 11-37 as anticipated by Soderland (U.S. 6,013,431). Applicant respectfully traverse this rejection for the reasons provided below.

A. The Claimed Subject Matter.

As an initial matter, Applicant notes that the pending claims are generally directed to methods of determining the presence or absence nucleic acids having a variation in a known nucleotide in a predetermined position of a nucleic acid of interest using primer extension techniques. The primer reaction mixture is formulated so that a labeled primer extension product will not form if the nucleic acid of interest contains the known nucleotide at the predetermined position. One way this primer extension reaction accomplishes this result is by using non-terminator nucleotides that are not complementary to the known nucleotide. Thus, if a nucleic acid of interest contains the known nucleotide in the predetermined position, a primer extension product cannot form because there are no nucleotides in the primer extension reaction mixture that are

complementary to the known nucleotide. Any primer extension that forms indicates that the nucleotide at the predetermined position was not the known nucleotide. The specific identity of the variant nucleotide at the predetermined position is irrelevant; the relevant aspect is whether or not the nucleotide at the predetermined position is the known nucleotide.

Another way the claimed subject matter detects nucleic acids having variations of a known nucleotide at a predetermined position in a nucleic acid of interest is by using terminator nucleotides in the primer extension reaction mixture that are complementary to the known nucleotide. As a result, if the nucleic acid of interest includes the known nucleotide in the predetermined position, the terminator nucleotide will be incorporated during the primer extension reaction and will prevent the subsequent incorporation of labeled non-terminator nucleotides.

B. Soderland Fails To Disclose Each Element Of The Claims.

For a proper rejection of a claim under 35 U.S.C. Section 102, the cited reference must disclose all elements of the claim. See, *e.g., E.I. du Pont de Nemours & Co. v. Phillips Petroleum Co.*, 849 F.2d 1430, 7 USPQ2d 1129 (Fed. Cir. 1988). Applicant respectfully submits that Soderland fails to disclose each element of the claims and therefore cannot anticipate the claims.

1. Claim 37

Claim 37 as amended provides:

> A method for detecting or quantifying the presence of a nucleic acid of interest having a variant of a known nucleotide base in a predetermined position of a known nucleic acid comprising:
>
> (a) obtaining a nucleic acid of interest having a target nucleotide base at the predetermined position in a template of the nucleic acid of interest;
>
> (b) preparing an unlabeled primer complementary to a sequence immediately upstream of the target nucleotide base;
>
> (c) treating ~~a~~ sample containing the nucleic acid of interest, if the nucleic acid is double-stranded, so as to obtain unpaired nucleotide bases spanning the predetermined position, or directly employing step (d) if the nucleic acid of interest is single-stranded;
>
> (d) annealing the primer from (b) with the nucleic acid of interest from (c) to obtain a primer-nucleic acid duplex;
>
> (e) mixing the primer-nucleic acid duplex from (d) with a primer extension reaction reagent comprising: (i) three types of non-terminator nucleotides that are not complementarily matched to the known nucleotide base in the predetermined position of the nucleic acid of interest, wherein at least one type

of the non-terminator nucleotide is labeled with a detectable marker; and optionally (ii) one type of terminator nucleotide that is complementarily matched to the known nucleotide base in the predetermined position of the nucleic acid of interest, wherein the terminator nucleotide is not labeled;

(f) extending the primer extension reaction by enzymatic or chemical means to form a labeled primer extension product comprising a plurality of labeled non-terminator nucleotides, wherein a labeled primer extension product does not form when the target nucleotide base in the predetermined position of the nucleic acid of interest is the same as the known target nucleotide base in the predetermined position of the known nucleic acid; and

(g) determining the presence of the nucleic acid of interest having the variant target nucleotide base at the predetermined position in the nucleic acid of interest by detecting the presence of the labeled primer extension product, wherein detecting the labeled primer extension product is not based on size.

As the final Office Action points out Soderland is directed to methods for determining the identity of a specific nucleotide at a defined site. In other words, Soderland is a sequencing method. The presently claimed subject matter is not a sequencing method because the identity of the nucleotide in the predetermined site is known. The pending claims are directed generally to detecting polynucleotides having a variant nucleotide at a predetermined position, not the identity of a potential variant. Thus, in one respect, the pending claims represent a binary system for determining the presence or absence of a known nucleotide in a predetermined position by the presence of absence of a detectable primer extension product.

Moreover, claim 37 requires a specific primer extension reaction mixture that is not disclosed in Soderland. Soderland fails to disclose a method for detecting or quantifying a nucleic acid having a variant of a known nucleotide at a predetermined position using a reaction mixture having: three types of non-terminator nucleotides that are not complementarily matched to the known nucleotide base in the predetermined position of the known nucleic acid, wherein at least one type of the non-terminator nucleotide is labeled with a detectable marker; and optionally one type of terminator nucleotide that is complementarily matched to the known nucleotide base in the predetermined position of the nucleic acid of interest, wherein the terminator nucleotide is not labeled.

Nothing in Soderland discloses using a primer extension reaction mixture that selectively omits the nucleotide complementary to a known nucleotide at the predetermined position. In fact, Soderland specifically teaches using a primer extension reaction mixture containing a

labeled known nucleotide so that the detection of a labeled primer extension product means that target nucleotide at the predetermined position is complementary to the labeled known nucleotide.

Additionally, Soderland fails to disclose using unlabeled ddNTPs that are complementary to a known nucleotide at a predetermined position so that labeled primer extension products will only form when there is a variation at the predetermined position. Although Soderland discloses the general use of labeled nucleotides in primer extension reactions, Soderland does not disclose formulating the primer extension reactions as presently claimed. Accordingly, Soderland cannot anticipate claim 37.

Moreover, step (f) of claim 37 recites that a labeled primer extension product does not form when the target nucleotide base in the predetermined position of the nucleic acid of interest is the same as the known target nucleotide base in the predetermined position of the known nucleic. Soderland fails to disclose a primer extension reaction wherein a primer extension product is not formed as recited in claim 37 because Soderland is directed to determining the identity of the nucleotide at the predetermined position. Accordingly, Soderland cannot anticipate claim 37 for at least this reason.

Because claims 2-9, and 11-36 depend from claim 37, the incorporate the limitations of claim 37. Accordingly, dependent claims 2-9 and 11-36 are not anticipated by Soderland for at least the reasons claim 37 is not anticipated by Soderland.

2. Claim 39

Claim 39 as amended provides:

> A method for detecting the presence of a nucleic acid having a variant of a known nucleotide base at a predetermined position, comprising:
>
> providing a nucleic acid having a target nucleotide base at the predetermined position;
>
> annealing a primer to the nucleic acid immediately 3' of the predetermined position;
>
> extending the primer in an extension reaction to form a labeled primer extension product using a reaction mixture comprising non-terminator nucleotides, wherein the non-terminator nucleotides are not complementarily matched to the known nucleotide base at the predetermined position and at least one non-terminator nucleotide is labeled with a detectable marker; and wherein a labeled primer extension product does not form when the target nucleotide base is

the same as the known nucleotide base at the predetermined position;

detecting the presence of the labeled primer extension product; and

correlating the presence of the labeled primer extension product with the presence of a nucleic acid having a variant of the known nucleotide base at the predetermined position in the nucleic acid.

As with claim 37, claim 39 requires a specifically formulated primer extension reaction mixture so that a labeled primer extension product does not form when the target nucleotide base is the same as the known nucleotide base at the predetermined position. This is accomplished by using a reaction mixture comprising non-terminator nucleotides, wherein the non-terminator nucleotides are not complementarily matched to the known nucleotide base at the predetermined position and at least one non-terminator nucleotide is labeled with a detectable marker. Soderland fails to disclose at least this element of claim 39, and therefor cannot anticipate the claim. Nor does Soderland disclose correlating the formation of a primer extension product with the presence of a polynucleotide having a variant of a known nucleotide in the predetermined position using the specifically formulated primer extension reaction mixture. As a result, Applicant respectfully submits that Soderland cannot anticipate claim 39.

3. Claim 40

Claim 40 as amended provides:

A method for detecting the presence of a nucleic acid having a known nucleotide at a predetermined position, comprising:

providing a nucleic acid sample having the nucleic acid with a target nucleotide at the predetermined position;

annealing a primer to the nucleic acid immediately 3' of the predetermined position;

extending the primer to form a labeled primer extension product using a reaction mixture comprising non-terminator nucleotides that are not complementarily matched to the known nucleotide base at the predetermined position and at least one non-terminator nucleotide is labeled with a detectable marker; and wherein a labeled primer extension product does not form when the identity of the target nucleotide base is the same as the known nucleotide base at the predetermined position; and

detecting the presence of the labeled primer extension product, wherein the absence of a detectable labeled primer extension product indicates the

presence of the known nucleotide base at the predetermined position in the nucleic acid.

Claim 40 also contains a limitation on the primer extension reaction mixture that is not disclosed by Soderland. In claim 40, the reaction mixture includes non-terminator nucleotides that are not complementarily matched to the known nucleotide base at the predetermined position and at least one non-terminator nucleotide is labeled with a detectable marker. By using this reaction a mixture, a labeled primer extension product does not form when the identity of the target nucleotide base is the same as the known nucleotide base at the predetermined position. Applicant respectfylly submits that Soderland does not disclose using a reaction mixture as presently claimed so that a labeled primer extension product does not form when the identity of the target nucleotide base is the same as the known nucleotide base at the predetermined position. Thus, Soderland cannot anticipate claim 40.

4. Claim 41

Claim 41 as amended provides:

A method for detecting or quantifying the presence of a known target nucleic acid in a sample by detecting a signal from a plurality of labeled nucleotides incorporated into a primer extension product comprising:

(a) selecting a nucleic acid having a target nucleotide base at a predetermined position in a template of a nucleic acid of interest;

(b) preparing an unlabeled primer complementary to a sequence immediately upstream of the target nucleotide base;

(c) treating a sample containing the nucleic acid of interest, if the nucleic acid is double-stranded, so as to obtain unpaired nucleotide bases spanning the predetermined position, or directly employing step (d) if the nucleic acid of interest is single-stranded;

(d) annealing the primer from (b) with the nucleic acid of interest from (c) to obtain a primer-nucleic acid duplex, wherein the target nucleotide base in the nucleic acid of interest is the first unpaired base immediately downstream of the 3' end of the primer;

(e) mixing the primer-nucleic acid duplex from (d) with a primer extension reaction reagent comprising: (i) non-terminator nucleotides that are not complementarily matched to the known target nucleotide base, wherein at least one type of the non-terminator nucleotide is labeled with a detectable marker; and optionally (ii) one type of terminator nucleotide that is complementarily matched

to the known target nucleotide base, wherein the terminator nucleotide is not labeled;

(f) extending the primer extension reaction by enzymatic or chemical means to form a labeled primer extension product comprising a plurality of labeled non-terminator nucleotides, wherein a labeled primer extension product does not form when the target nucleotide base is the known nucleotide; and

(g) wherein the absence of a detectable primer extension product indicates the presence of the known target nucleotide at the predetermined position in the nucleic acid of interest.

Claim 41 also requires a primer reaction mixture that is not disclosed by Soderland. The recited primer extension reaction mixture includes non-terminator nucleotides that are not complementarily matched to the known target nucleotide base, wherein at least one type of the non-terminator nucleotide is labeled with a detectable marker; and optionally one type of terminator nucleotide that is complementarily matched to the known target nucleotide base, wherein the terminator nucleotide is not labeled. The reaction mixture is formulated so that the absence of a detectable primer extension product indicates the presence of the known target nucleotide at the predetermined position in the nucleic acid of interest. Applicant respectfully submits that Soderland fails to disclose at least this element. Accordingly, Soderland cannot anticipate claim 41.

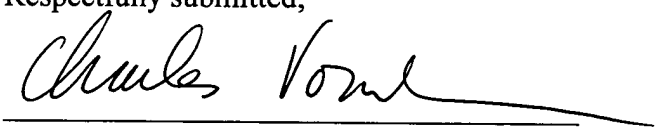
3. Information Disclosure Statement

Applicant notes that previously submitted references AJ, L, M, and P filed on July 21, 2003, were not considered because the references were not with the application when forward to the Examiner. Applicant encloses copies of these references and a supplemental Information Disclosure Form listing these references and respectfully requests that the Examiner consider them. Applicant believes no fee is due with this Information Disclosure Statement because the references were previously submitted; however, in the event the Examiner determines a fee is required, the Commissioner is authorized to charge the requisite fee to Deposit Account No. 20-0778.

CONCLUSION

In light of the foregoing amendments and remarks Applicant respectfully submits that all rejections have been traversed or rendered moot, and the now pending claims 1-3, 5, 8, 9, 11-37, and 39-41 are in condition for allowance. Favorable reconsideration and allowance of the present application and all pending claims are hereby courteously requested. If, in the opinion of the Examiner, a telephonic conference would expedite the examination of this matter, the Examiner is invited to call the undersigned agent at (770) 933-9500.

Respectfully submitted,



Charles Vorndran, Reg. No. 45,415

THOMAS, KAYDEN,
HORSTEMEYER & RISLEY, L.L.P.
Suite 1750
100 Galleria Parkway N.W.
Atlanta, Georgia 30339
(770) 933-9500